

**EFFECTS OF SURFACE ACTIVE AGENTS
ON MARINE MUSSELS AND FISH**

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The thesis is based on the following papers:

- I Biological effects of surface active agents on marine animals.
M. Swedmark; B. Braaten; E. Emanuelsson and Å. Granmo. Mar. Biol. 9, 183 - 201 (1971)
- II Effects of oil dispersants and oil emulsions on marine animals.
M. Swedmark; Å. Granmo and S. Kollberg. Wat. Res. 7, 1649 - 1672 (1973)
- III Effects on marine animals of long-term exposure to surfactants.
Å. Granmo; I. Berg; S. Kollberg and M. Swedmark (in manuscript)
- IV Development and growth of eggs and larvae of *Mytilus edulis* L. exposed to a linear dodecylbenzensulphonate, LAS. Mar. Biol. 15, 356 - 358 (1972)
- V Effects on fertilization and development of the common mussel (*Mytilus edulis* L.) at long-term exposure to a nonionic surfactant.
Å. Granmo and G. Jørgensen. Mar. Biol. (in press)
- VI Uptake pathways and elimination of a nonionic surfactant in cod (*Gadus morrhua* L.) Å. Granmo and S. Kollberg. (in manuscript)
- VII Tissue-swelling in *Mytilus edulis* L. induced by exposure to a nonionic surface active agent. B. Braaten; Å. Granmo and R. Lange. Norw. J. Zool. 20, 137 - 140 (1972)

In the dissertation these papers are referred to by the Roman figures given above.

INTRODUCTION

During the 1950's synthetic surface active agents (surfactants) replaced the classical soaps in detergents. The world consumption of these substances amounted in 1969 to about 8 million tons, and thus constitute an important pollutant in the aquatic environment. Much work has been done to determine their toxicity to fish, but little was known about the tolerance of marine invertebrates to surfactants, most of the available information being concerned with the toxicity of oil-dispersants (Corner *et al.*, 1968; Portmann and Connor, 1968; Bellan *et al.*, 1969, Perkins, 1970).

The present investigations were undertaken as a comparative study of the effects of certain surfactants now used in the manufacturing of detergents and oil-dispersants. Both acute and chronic toxicity of surfactants were investigated (I, III) as also acute toxicity of oil-dispersants (II). Further, the sensitivity of young stages of mussels was studied (IV, V). Finally, efforts were made to contribute to the explanation of the action of surfactants, by studying the effects of hydration in exposed mussels (VII) as also the uptake pathways, distribution in the body and elimination in cod exposed to surfactants (VI).

MATERIAL AND METHODS

SURFACTANTS

In all, 3 anionic and 2 nonionic surfactants were chosen for testing:
tetrapropylene benzene sulphonate, ABS (I)
linear dodecylbenzene sulphonate, LAS (I, III, V)
lauryl ether sulphate, LES 3 EO (I)
nonylphenol ethoxylate, NP 10 EO (I, III, VI, VII)
tallow alcohol ethoxylate, TAE 10 EO (I, III, V)

TEST ANIMALS

In all investigations except no. VI, different species of bivalves caught in the Gullmar Fjord were used.

Mytilus edulis (I, II, III, IV, V, VII)

Cardium edule (I, II, III)

Mya arenaria (I)
Ostrea edulis (I)
Astarte sulcata and *A. montagui* (I)
Pecten maximus (I)
Pecten opercularis (II, III)
In investigation VI, cod (*Gadus morrhua*) was used.

EXPERIMENTAL CONDITIONS

As a rule a continuous flow system was used as described in I, by Granmo and Kollberg (1972) and Berg and Granmo (in press). In the studies of mussel larvae (IV, V) a static system was used, as also in the tests concerning the action of surfactants (IV, VII).

In all experiments the salinity was kept at 32 - 34 ‰, and temperature constant at two or three defined levels except in the long-term testing where deep water from the Fjord was used.

BIOLOGICAL FUNCTIONS STUDIED

Short-term tests were run for 96 h with a subsequent recovery period in clean sea water. Mortality and survival times were continuously registered. The median lethal concentration (LS 50) was obtained by graphic interpolation on the cumulative percentage mortality curve plotted against exposure time.

Sublethal and chronic effects were studied in both the short-term and long-term tests. Different functions such as heart beat frequency, byssal activity, shell-closure ability, siphon retraction in response to prodding, burrowing activity and growth were observed.

Fertilization and development of different larval stages of common mussels were studied when exposed to certain surfactants (IV, V).

The uptake and elimination of a labelled surfactant, NP 10 EO, was studied in cod (VI). The amount of the surfactant was analyzed by the scintillation counting method in different tissues from the fish.

In common mussels exposed to a nonionic surfactant the water in the large adductor muscles were determined from the difference between the weight before and after drying at 105 °C for 24 h (VII).

RESULTS

ACUTE TOXICITY (I, II)

The bivalves were found to be less susceptible to surfactants than fishes but more susceptible than crustaceans. Within the group of bivalves more active species such as *Pecten* and *Cardium* were found more sensitive than less active species as *Mya* and *Mytilus*. In general nonionic surfactants proved to be more toxic than anionic ones, and "hard" types (ABS, NP 10 EO) caused more pronounced effects than "soft" ones (LAS, LES 3 EO, TAE 10 EO) in the concentrations used, while the opposite was found in the fishes.

The toxicity of the surfactants for the mussels, expressed in 96 h LC 50 values, lay within the range of < 5 to > 100 ppm. The effects were more pronounced at higher temperatures.

Ability to recover normal behavior after exposure decreased with increasing concentration and time, and ceased earlier in anionic than in nonionic surfactants.

Sublethal effects on different functions were observed for all the surfactants. Sensitive functions were byssal activity in *Mytilus*, siphon retraction in *Cardium* and *Mya*, and the burrowing activity in *Cardium* and *Astarte* (two species). A general pattern appeared, initial increased activity (shell-closure) being followed by inactivation, immobilization and death.

Also for oil-dispersants and oil-emulsions (II) bivalves were found to be most sensitive. This is accentuated by the delayed reactions, typical for bivalves, which continue during the period in clean water. A probable explanation may be their high rate of accumulation. The reactions of the species to the different dispersants varied greatly in strength.

CHRONIC TOXICITY (III)

The toxicity of the surfactants to the bivalves increased at prolonged exposure time. Further, the differences in acute toxicity between various types of surfactants and species decreased at long-term exposures.

The mortality rate generally increased with rising temperature. A similar pattern as in short-term tests was observed though the phases were more extended and often incomplete.

Intermittent dosage showed less serious effects for the animals compared to constant dosage, particularly evident with respect to mortality and growth in juvenile common mussels.

EFFECTS ON DEVELOPMENTAL STAGES OF COMMON MUSSELS (IV, V)

The fertilization and growth of eggs and larvae of *Mytilus* were studied under the influence of the anionic LAS. A reduction of the fertility was obtained in concentrations as low as 0.05 ppm, i.e. less than 1/2000 of the 96 h LC 50 value for the adult mussel (IV).

Gametes from mussels, long-term exposed in TAE 10 EO, were found to be more sensitive than those from the corresponding control; they displayed an inhibited or delayed development (V).

ACTION OF THE SURFACTANTS (VI, VII)

The penetration, uptake and elimination of the nonionic NP 10 EO in cod was found to be a rapid process. The principal site of penetration was found to be the gills. Further, high amount of NP 10 EO found in blood after 5 min. exposure revealed that the blood is the chief vehicle of transportation to the various tissues where deposition occurs. High concentrations were found in liver and gall bladder. After 24 h recovery in clean sea water the residues in different organs had decreased by 60 %.

Mytilus individuals exposed to NP 10 EO showed an increased hydration of the muscle tissues. The amount of the tissue-swelling varied as a function of the exposure time and the concentration of the surfactant. The susceptibility of *Mytilus* to the surfactant, measured as tissue-swelling, varied during the year. It was less in the spawning season than in other parts of the year. It is concluded that the swelling of the tissue is due to damage to the cell membranes.

CONCLUSIONS

Sublethal effects on biological functions such as those observed are of considerable importance for the survival of the animals in their natural environment. The increased activity at exposure to surfactants is interpreted as an avoidance reaction expressed as escape movements in mobile species such as *Pecten* (Thomas and Gruffydd, 1971) or protective behaviours, such as shell-closure in sessile bivalves. Evasive reactions seem to be especially well developed in littoral species, naturally adapted to sudden variations of their environment.

In addition to the direct toxic effects of surfactants on the animals they also affect the permeability of cell membranes allowing other substances, for example heavy metals, to enter the cells more easily. Then they may give synergistic effects in recipients depending on the other pollutants present.

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REFERENCES

- Bellan, G, F. Caruelle, P. Foret-Montardo, R. A. Kaim-Malka et K. Leung Tack (1969) Contribution à l'étude de différents facteurs physicochimiques polluants sur les organismes marins. I. Action des détergents sur la polychete *Scolecopsis fuliginosa* (note préliminaire). Tethys 1, 367 - 374.
- Berg, I. and Å. Granmo (in press) Procedures for toxicity testing in a continuous flow system. FAO, FIR/TPLR/73/M6, 9 pp.
- Corner E. D. S. , A. J. Southward and E. C. Southward (1968) Toxicity of oil-spill removers ("detergents") to marine life: an assessment using the intertidal barnacle *Elminius modestus*. J. Mar. biol. Ass. U.K. 48, 29 - 47.
- Granmo, Å. and S. O. Kollberg (1972) A new simple water flow system for accurate continuous flow tests. Wat. Res. 6, 1597 - 1599.
- Perkins, E. J. (1970) Some effects of detergents in the marine environment. Chemy. Ind. 3, 14 - 22.
- Portmann J. E. and P. M. Connor (1968) The toxicity of several oil-spill removers to some species of fish and shellfish. Mar. Biol. 1, 322 - 329.
- Thomas, G. E. and Ll. D. Gruffydd (1971) The types of escape reactions elicited in the scallop *Pecten maximus* by selected sea-star species. Mar. Biol. 10, 87 - 93.

